

=> file medline
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SINCE FILE	TOTAL
ENTRY	SESSION
0.15	0.15

FILE 'MEDLINE' ENTERED AT 16:00:04 ON 14 DEC 2001

FILE LAST UPDATED: 13 DEC 2001 (20011213/UP). FILE COVERS 1958 TO DATE.

On April 22, 2001, MEDLINE was reloaded. See HELP RLOAD for details.

MEDLINE now contains IN-PROCESS records. See HELP CONTENT for details.

MEDLINE is now updated 4 times per week. A new current-awareness alert frequency (EVERYUPDATE) is available. See HELP UPDATE for more information.

MEDLINE thesauri in the /CN, /CT, and /MN fields incorporate the MeSH 2001 vocabulary. Enter HELP THESAURUS for details.

The OLDMEDLINE file segment now contains data from 1958 through 1965. Enter HELP CONTENT for details.

Left, right, and simultaneous left and right truncation are available in the Basic Index. See HELP SFIELDS for details.

THIS FILE CONTAINS CAS REGISTRY NUMBERS FOR EASY AND ACCURATE SUBSTANCE IDENTIFICATION.

=> s (Donoho, G? or Donoho G?)/au
 6 DONOHO, G?/AU
 6 DONOHO G?/AU
 L1 6 (DONOHO, G? OR DONOHO G?)/AU

=> d L1 1-6 ti au so abs

L1 ANSWER 1 OF 6 MEDLINE
 TI Cells deleted for Brca2 COOH terminus exhibit hypersensitivity to gamma-radiation and premature senescence.
 AU Morimatsu M; Donoho G; Hasty P
 SO CANCER RESEARCH, (1998 Aug 1) 58 (15) 3441-7.
 Journal code: CNF; 2984705R. ISSN: 0008-5472.
 AB The putative Brca2-MmRad51 interaction is analyzed in mouse cells deleted for the COOH terminus of Brca2 (amino acids 3140-3328), which contains a region that associates with MmRad51 by yeast two-hybrid. These cells are hypersensitive to gamma-radiation (suggesting defective recombinational repair) but not UV light (suggesting intact nucleotide excision repair) and maintain the G1-S and G2-M checkpoints after exposure to gamma-irradiation. Cells deleted for the COOH terminus of Brca2 progress through the cell cycle at a similar rate as wild-type cells but undergo senescence more rapidly. These data support the hypothesis that deletion of Brca2 stimulates cancer by defective MmRad51-mediated DNA repair and not by defective cell cycle regulation.

L1 ANSWER 2 OF 6 MEDLINE
 TI Analysis of gene targeting and intrachromosomal homologous recombination stimulated by genomic double-strand breaks in mouse embryonic stem cells.
 AU Donoho G; Jasinska M; Berg P
 SO MOLECULAR AND CELLULAR BIOLOGY, (1998 Jul) 18 (7) 4070-8.
 Journal code: NGY; 8109087. ISSN: 0270-7306.
 AB To investigate the effects of in vivo genomic DNA double-strand breaks on the efficiency and mechanisms of gene targeting in mouse embryonic stem cells, we have used a series of insertion and replacement vectors carrying two, one, or no genomic sites for the rare-cutting endonuclease I-SceI. These vectors were introduced into the hypoxanthine phosphoribosyltransferase (hprt) gene to produce substrates for gene-targeting (plasmid-to-chromosome) or intrachromosomal (direct repeat)

homologous recombination. Recombination at the hprt locus is markedly increased following transfection with an I-SceI expression plasmid and a homologous donor plasmid (if needed). The frequency of gene targeting in clones with an I-SceI site attains a value of 1%, 5,000-fold higher than that in clones with no I-SceI site. The use of silent restriction site polymorphisms indicates that the frequencies with which donor plasmid sequences replace the target chromosomal sequences decrease with distance from the genomic break site. The frequency of intrachromosomal recombination reaches a value of 3.1%, 120-fold higher than background spontaneous recombination. Because palindromic insertions were used as polymorphic markers, a significant number of recombinants exhibit distinct genotypic sectoring among daughter cells from a single clone, suggesting the existence of heteroduplex DNA in the original recombination product.

L1 ANSWER 3 OF 6 MEDLINE

TI A polyoma-based episomal vector efficiently expresses exogenous genes in mouse embryonic stem cells.

AU Camenisch G; Gruber M; **Donoho G**; Van Sloun P; Wenger R H;
Gassmann M

SO NUCLEIC ACIDS RESEARCH, (1996 Oct 1) 24 (19) 3707-13.
Journal code: O8L; 0411011. ISSN: 0305-1048.

AB We describe the ability of novel episomally maintained vectors to efficiently promote gene expression in embryonic stem (ES) cells as well as in established mouse cell lines. Extrachromosomal maintenance of our vectors is based on the presence of polyoma virus DNA sequences, including the origin of replication harboring a mutant enhancer (PyF101), and a modified version of the polyoma early region (LT20) encoding the large T antigen only. Reporter gene expression from such extrachromosomally replicating vectors was approximately 10-fold higher than expression from replication-incompetent control plasmids. After transfection of different ES cell lines, the polyoma virus-derived plasmid variant pMGD20neo (7.2 kb) was maintained episomally in 16% of the G418-resistant clones. No chromosomal integration of pMGD20neo vector DNA was detected in ES cells that contained episomal vector DNA even after long term passage. The vector's replication ability was not altered after insertion of up to 10 kb hprt gene fragments. Besides undifferentiated ES cells, the polyoma-based vectors were also maintained extrachromosomally in differentiating ES cells and embryoid bodies as well as in established mouse cell lines.

L1 ANSWER 4 OF 6 MEDLINE

TI Maintenance of an extrachromosomal plasmid vector in mouse embryonic stem cells.

AU Gassmann M; **Donoho G**; Berg P

SO PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1995 Feb 28) 92 (5) 1292-6.

Journal code: PV3; 7505876. ISSN: 0027-8424.

AB We have constructed and characterized a polyoma virus-based plasmid that is maintained as an autonomously replicating extrachromosomal element (episome) in mouse embryonic stem (ES) cells. Plasmid pMGD20neo contains the polyoma origin of replication harboring a mutated enhancer (PyF101), a modified polyoma early region that encodes the large tumor (T) antigen only, and a gene that confers resistance to G418 (neo). After transfection, the plasmid replicates in ES cells and is maintained as an extrachromosomal element in 15% of G418-resistant clones. Integration of the plasmid DNA is undetectable for at least 28 cell generations. In one clone, the transfected DNA persists unaltered as an episome at 10-30 copies per cell for at least 74 cell generations in the presence of G418. Cells that maintain the autonomously replicating plasmid can efficiently replicate and maintain a second plasmid that carries the polyoma origin of replication. Independent vector-containing ES cell lines showed no significant alteration of the karyotype, and two cell lines yielded several chimeric animals when introduced into blastocysts, suggesting that the presence of an episomal element and expression of polyoma large T do not eliminate the ES cells' ability to populate an embryo. This system offers an efficient means for manipulating and analyzing various aspects of gene expression in ES cells.

L1 ANSWER 5 OF 6 MEDLINE
TI Effect of ethanol and low-temperature culture on expression of soybean
lipoxygenase L-1 in Escherichia coli.
AU Steczko J; **Donoho G A**; Dixon J E; Sugimoto T; Axelrod B
SO PROTEIN EXPRESSION AND PURIFICATION, (1991 Apr-Jun) 2 (2-3) 221-7.
Journal code: BJV; 9101496. ISSN: 1046-5928.
AB We have constructed a full-length cDNA that encodes soybean seed
lipoxygenase L-1 and have expressed it in Escherichia coli. This gene was
inserted into a pT7-7 expression vector, containing the T7 RNA polymerase
promoter. *E. coli*, strain BL21 (DE3), which carries the T7 promoter in its
genome, was transfected with the plasmid. Expression of this gene when the
cells were cultured at 37 degrees C yielded polypeptide that was
recognized by anti-L-1 antibody, but had very little lipoxygenase
activity. Yields of active enzyme were markedly increased when cells were
cultured at 15-20 degrees C. When ethanol, which has been reported to be
an excellent elicitor of heat-shock proteins in *E. coli*, was also present
at a level of 3% the yield was further increased by 40%. Under optimum
conditions 22-30 mg of soluble active enzyme was obtained per liter of
culture.

L1 ANSWER 6 OF 6 MEDLINE
TI Conserved histidine residues in soybean lipoxygenase: functional
consequences of their replacement.
AU Steczko J; **Donoho G P**; Clemens J C; Dixon J E; Axelrod B
SO BIOCHEMISTRY, (1992 Apr 28) 31 (16) 4053-7.
Journal code: AOG; 0370623. ISSN: 0006-2960.
AB Sequences of 13 lipoxygenases from various plant and mammalian species,
thus far reported, display a motif of 38 amino acid residues which
includes 5 conserved histidines and a 6th histidine about 160 residues
downstream. These residues occur at positions 494, 499, 504, 522, 531, and
690 in soybean lipoxygenase isozyme L-1. Since the participation of iron
in the lipoxygenase reaction has been established and existing evidence
based on Mossbauer and EXAFS spectroscopy suggests that histidines may be
involved in iron binding, the effect of the above residues has been
examined in soybean lipoxygenase L-1. Six singly mutated lipoxygenases
have been produced in which each of the His residues has been replaced
with glutamine. Two additional mutants have been constructed wherein the
codons for His-494 and His-504 have been replaced by serine codons. All of
the mutant lipoxygenases, which were obtained by expression in *Escherichia*
coli, have mobilities identical to that of the wild-type enzyme on
denaturing gel electrophoresis and respond to lipoxygenase antibodies. The
mutated proteins H499Q, H504Q, H504S, and H690Q are virtually inactive,
while H522Q has about 1% of the wild-type activity. H494Q, H494S, and
H531Q are about 37%, 8%, and 20% as active as the wild type, respectively.
His-517 is conserved in the several lipoxygenase isozymes but not in the
animal isozymes. The mutant H517Q has about 33% of the wild-type activity.
The inactive mutants, H499Q, H504Q, H504S, and H690Q, become insoluble
when heated for 3 min at 65 degrees C, as does H522Q. The other mutants
and the wild-type are stable under these conditions. (ABSTRACT TRUNCATED AT
250 WORDS)

=> s (Hilbun, E? or Hilbun E?)/au
0 HILBUN, E?/AU
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L2 0 (HILBUN, E? OR HILBUN E?)/AU